Atty. Docket No.: 051058-029000 (Formerly NUCL-001/01) **PATENT**

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Application of:	Satishchandran, C	Examiner:	Chong, Kimberly
Serial No.:	10/009,134		
Filed:	October 20, 2002	Group Art Unit:	1635
Entitled:	METHODS AND COMPOSITIONS FOR INHIBITING THE FUNCTION OF POLYNUCLEOTIDE SEQUENCE		
		Conf. No.:	2937

Mail Stop RCE
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION OF DR. CATHERINE PACHUK UNDER 37 C.F.R. 1.132

I declare:

1. I, Catherine Pachuk, hold the position of Senior Director at the Pfizer Research Technology Center of Pfizer Pharmaceuticals, Cambridge, MA.

2. I hold a Ph.D. Degree in Molecular Biology from The University of Pennsylvania. I am an author on more than 20 peer-reviewed literature publications. A copy of my Curriculum Vitae is attached.

3. I am an inventor on the above-noted U.S. patent application.

4. I have read the Office Action issued November 18, 2009 in the above-noted patent application, and I understand that the Examiner has rejected claims 107-114, 116-136, 138-140, 142-145, 147, 157-167 and 172-174 as being obvious in view of the teachings of Werther et al. (U.S. Patent No. 5,929,040), Fire et al. (U.S. Patent No. 6,506,559), Heifetz et al. (WO99/61631), Calbretta et al (U.S. Patent No. 5,734,039), Taira et al. (U.S. Patent No. 5,500,357), and Thompson et al. (U.S. Patent No. 6,146,886). I also understand that the Examiner has also rejected the above-noted claims as being obvious in view of Taira et al. (U.S.

Patent No. 5,500,357), Fire et al. (U.S. Patent No. 6,506,559) and Thompson et al. (U.S. Patent No. 6,146,886).

5. In particular, I understand that the Examiner is arguing that it would have been obvious to take elements from the ribozyme and antisense arts and apply such elements to the field of post-transcriptional gene silencing using double stranded RNA. The Examiner indicates that this argument is based on a similar mechanism of action among ribozymes, antisense and dsRNA, mainly that each utilize a single stranded antisense sequence for binding to the target RNA. The Office Action states at the paragraph bridging pages 4-5:

> "Moreover, it is well known in the art that ribozymes, antisense molecules and siRNA all cleave the target RNA by different mechanisms however it **is also well known in the art that each of the molecules *require a single stranded antisense sequence* that recognizes and binds to the target RNA to mediate cleavage of the target RNA** and inhibition of expression which is the common purpose of each of the molecules." (Emphases added)

6. I disagree with the statement outlined above. It is now well known in the field that the *mechanism of association* with a target RNA is different between antisense/ribozymes and siRNA. Furthermore, the field of post-transcriptional gene silencing using double stranded RNA (dsRNA) molecules was in its infancy at the time of filing and the mechanism of action of a double-stranded RNA effector molecule in mediating target cleavage of an RNA was not yet fully understood. In particular, it was not known that dsRNAs bind to the target mRNA using a single stranded antisense sequence. Further, the involvement of the RISC complex for processing small dsRNA effector molecules was not yet known. It was not until the RISC complex was implicated in the unwinding and cleavage of dsRNA molecules that it was first hypothesized that the antisense strand of a dsRNA molecule binds to the target RNA to mediate cleavage (and this is via RISC) (Elbashir et al., 2001 (Exhibit A); Nykanen et al. (Exhibit B), 2001; and Martinez et al., 2002 (Exhibit C)). At the time that the present application was filed, it was not yet known in the art that dsRNA molecules acted through a mechanism whereby the antisense sequence binds to a complementary region of a target RNA to mediate cleavage. It follows then that the Examiner's arguments are based on parts

of what are known in the art, at best, at a time after the present application was filed, and at worst, at the *present* time, rather than what was known in the art at the time of filing.

7. In view of the above, it is my opinion that those skilled in the art as of April 29, 1999 (i.e., the earliest priority date of the present application) did not know, and could not have known, that a single strand of a dsRNA molecule binds to a target RNA to mediate cleavage. It was not clear how a double stranded siRNA could bind to a single strand RNA. This is a big difference between antisense RNA and ribozymes which each have single stranded regions that enable binding. Simple melting of the siRNA could not explain this, because, for many siRNAs, the Tms were too high for duplex melting to be a plausible explanation.. This knowledge that an siRNA can bind to a target RNA was not available in the art until, at the earliest, the 2001 publication of Nykanen et al. (*Cell* 107:309-321; Exhibit A) that implicated the role of RISC in unwinding of a dsRNA molecule. Even then, it was not clearly shown that RISC is associated with an *antisense* strand of a dsRNA until a study published by Martinez et al. (2002) (Cell, volume 110: 563-574; Exhibit C).

8. In my opinion and in view of the above, one of skill in the art at the time of the invention would not have recognized that ribozymes, antisense and dsRNA each require a single strand of RNA that binds to a complementary region of a target RNA to effect target cleavage. As such, one of skill in the art would not have been motivated to take elements known to work in the ribozyme and antisense literature and apply them to molecules for dsRNA mediated inhibition of gene expression. In particular, one of skill in the art would not have combined the teachings of the Werther et al., Fire et al., Heifetz et al., Calabretta et al., Taira et al., and Thompson et al. references to arrive at the presently claimed invention as proposed by the Examiner. Further, it was not known that the elements of antisense and/or ribozymes for mediating inhibition of gene expression could be applied together with, or would be amenable to, the generation or activity of siRNA molecules from precursor RNAs.

9. In my opinion and in view of the above, the Examiner has reviewed the present invention with a substantial bias to the knowledge available in the art at the present time and has not taken into account the knowledge understood in the art at the time of filing, particularly with regard to the mechanism of action of a dsRNA molecule. That is, contrary to the Examiner's assertions in

the Office Action, it was not well known in the art at the time that each of antisense-, ribozyme- and dsRNA-mediated inhibition "require a single stranded antisense sequence that recognizes and binds to the target RNA to mediate cleavage of the target RNA." In my opinion then, the reasoning regarding the obviousness conclusion is based on an incorrect assumption regarding what was known in the art at the time.

10. I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patents issued thereon.

8-25-10

Date

Catherine Pachuk

Catherine Pachuk, Ph.D.